

AMENDMENT TO THE SPECIFICATION

Please amend paragraph [0019] as follows:

[0019]

[Fig. 1]

It is a figure that shows the results of sandwich ELISA to αs1 caseins at various states by using 2 types of anti-αs1 casein MAbs of the present invention (milk allergen).

[Fig. 2]

It is a figure that shows the difference of component protein of flour-αs1 casein that is recognized by Pas1CN1 and Pas1CN2 of the present invention (milk allergen).

[Fig. 3]

It is a figure that shows the reactivity of PLG2 and PLG1 against various β-lactoglobulins by sandwich ELISA of the present invention (milk allergen).

[Fig. 4]

It is a figure that shows the reactivity of PLG2 and PLG3 against various β-lactoglobulins by sandwich ELISA of the present invention (milk allergen).

[Fig. 5]

It is a figure that shows the reactivity against native lactoglobulins in a MAb-mixed system by sandwich ELISA of the present invention (milk allergen).

[Fig. 6]

It is a figure that shows the reactivity against urea-treated lactoglobulins in a MAb-mixed system by sandwich ELISA of the present invention (milk allergen).

[Fig. 7]

It is a figure that shows the reactivity of anti-ovalbumin MAbs against each serial dilution in Test 1 of the present invention (albumen allergen).

[Fig. 8]

It is a figure that shows the reactivity of anti-ovalbumin MAbs against each serial dilution in Test 2 of the present invention (albumen allergen).

[Fig. 9]

It is a figure that shows the reactivity of anti-ovalbumin MAbs against each serial dilution in Test 3 of the present invention (albumen allergen).

[Fig. 10]

It is a figure that shows the reactivity of PNOM1 and PNOM2 against denatured/native ovomucoid by sandwich ELISA of the present invention (albumen allergen).

[Fig. 11]

It is a figure that shows the reactivity of PDOM1 and PDOM2 against denatured/native ovomucoid by sandwich ELISA of the present invention (albumen allergen).

[Fig. 12]

It is a figure that shows the reactivity of [[PDOM2]] PNOM2 and PDOM2, and PNOM1 and PDOM1 against denatured/native ovomucoid by sandwich ELISA of the present invention (albumen allergen).

[Fig. 13]

It is a figure that shows the results of sandwich ELISA against gliadin in various states, using 2 types of anti-gliadin MAbs of the present invention (flour allergen).

[Fig. 14]

It is a figure that shows the difference of constitutive protein of flour gliadin recognized by PGL1 and PGL2 of the present invention (flour allergen).

[Fig. 15]

It is a figure that shows the reactivity of PBW2 and PBW3 against various buckwheat crude proteins by sandwich ELISA of the present invention (buckwheat allergen).

[Fig. 16]

It is a figure that shows the reactivity of PBW1 and PBW2 against various buckwheat crude proteins by sandwich ELISA of the present invention (buckwheat allergen).

[Fig. 17]

It is a figure that shows the reactivity of MAb-mixed system of PBW1, PBW2 and PBW3 against native buckwheat crude proteins by sandwich ELISA of the present invention (buckwheat allergen).

[Fig. 18]

It is a figure that shows the reactivity of MAb-mixed system of PBW1, PBW2 and PBW3 against denatured buckwheat crude proteins by sandwich ELISA of the present invention (buckwheat allergen).

[Fig. 19]

It is a figure that shows the reactivity of PAh1-1 and PAh1-2 against various peanut crude proteins by sandwich ELISA of the present invention (peanut allergen).

[Fig. 20]

It is a figure that shows the reactivity of PAh1-2 and PAh1-3 against various peanut crude proteins by sandwich ELISA of the present invention (peanut allergen).

[Fig. 21]

It is a figure that shows the reactivity of MAb-mixed system of PAh1-1, PAh1-2 and PAh1-3 against native peanut crude proteins by sandwich ELISA of the present invention (peanut allergen).

[Fig. 22]

It is a figure that shows the reactivity of MAb-mixed system of PAh1-1, PAh1-2 and PAh1-3 against denatured peanut crude proteins by sandwich ELISA of the present invention (peanut allergen).

Please amend paragraph [0026] as follows:

[0026]

As the above anti-ovalbumin monoclonal antibodies, anti-ovalbumin monoclonal antibodies recognizing a native ovalbumin and/or a reduced carboxymethylated ovalbumin are preferable. Specifically, the anti-ovalbumin monoclonal antibody PNOA1 produced by hybridoma (FERM ABP-10265), the anti-ovalbumin monoclonal antibody PNOA2 produced by hybridoma (FERM ABP-10266), the anti ovalbumin monoclonal antibody PDOA1 produced by hybridoma (FERM ABP-10275), the anti ovalbumin monoclonal antibody PDOA2 produced by hybridoma (FERM ABP-10276) etc. can be preferably exemplified. Further, by using the combination of anti-native ovalbumin monoclonal antibodies such as PNOA1 and PNOA2, and anti denatured ovalbumin monoclonal antibodies such as PDOA1 and PDOA2, or especially by combining anti-native ovalbumin monoclonal antibodies such as PNOA1 and PNOA2 with anti-native denatured ovalbumin monoclonal antibodies such as PDOA1 and PDOA2, sandwich ELISA or immunochromatography can be performed more advantageously. For example, by using these antibodies, native ovalbumin and/or denatured ovalbumin in foods can be analyzed qualitatively and quantitatively even at a concentration between 1.0 to 10.0 ppb by sandwich ELISA.

Please amend paragraph [0038] as follows:

[0038]

Anti-food allergen MAb-producing hybridomas can be produced by, for example, immunizing a BALB/c mouse by using native and/or denatured food allergens, performing cell fusion of antibody-producing cells of the immunized mouse and mouse myeloma myeloma cells by common methods, and screening by immunofluorescent staining patterns. The above antibody-producing cells include, for example, spleen cells, lymph node cells and B-lymphocytes obtained from immunized animals that have been administered with native and/or denatured food allergens or a composition containing the same. As animals to immunize, mice, rats, rabbits and horses can be exemplified. Immunization is performed by, for example, administering native and/or denatured food allergens directly or with an appropriate adjuvant to an animal, subcutaneously, intramuscularly or intraperitoneally, 1 or 2 times per month, for 1 to 6 months. Separation of antibody-producing cells is performed by collecting from the immunized animals, 2 to 4 days after the final immunization. As myeloma cells, those derived from mice or rats can be used. It is preferable that antibody-producing cells and myeloma cells are from the animals of the same species.

Please amend paragraph [0040] as follows:

[0040]

Labeled substances used for labeled antibody preparation are not particularly limited as long as it is a labeled substance that can induce a signal which can be detected alone or by reacting with other substances. Enzymes, fluorescent substances, chemical photosubstances, radioactive substances and gold colloids can be used. Enzymes include peroxidase, alkaline phosphatase,  $\beta$ -D-galactosidase, glucose oxydase, glucose-6-phosphate dehydrogenase, alcohol dehydrogenase, malate dehydrogenase, penicillinase, catalase, apoglucose oxidase, urease, luciferase or acetyl cholinesterase. Fluorescent substances include fluorseeine fluorescein isothiocyanate, phycobiliprotein, rare-earth metal chilate, dansylchloride or tetramethylrodamine isothiocyanate. Photosubstances include luminols, dioxetanes, acridinium salts. Radioactive

substances include  $^3\text{H}$ ,  $^{14}\text{C}$ ,  $^{125}\text{I}$  or  $^{131}\text{I}$ . When a labeled substance is an enzyme, substrates can be used to measure its activity as well as coloring agents, fluorescent agents, radioagents, according to need.

Please amend paragraph [0099] as follows:

[0099]

5) Cloning by limiting dilution

Presence of hybridomas provided as primary antibodies for ELISA and producing anti-OA antibodies was examined in the culture supernatant of each well of cell culture plate.

Hybridomas which tested positive against  $[[\alpha\text{CN}]] \alpha\text{OA}$  by ELISA were transferred to a 96-well cell culture plate (Becton Dickinson) and cloned by limiting dilution to 0.9 cell/well.

Meanwhile, as feeder cells, thymocytes of 4-weeks old BALB/c mouse were added to each well of the 96-well cell culture plate, to  $5 \times 10^6$  cells/well. RPMI 1640 media containing 10% of bovine fetal serum, 40 mM of 2-mercaptoethanol, 100 U/ml of penicillin, 100 g/ml of streptomycin were used for culturing cloned hybridomas.

Please amend the second occurrence of paragraph [0101] to read as follows:

~~[0101]~~ [0102]

8) Characteristics of MAbs, classes and subclasses of MAbs

Solid-phase method and liquid-phase method were used to determine the characteristics of anti-OAMAbs. As a solid-phase method, a method comprising the steps of fixing NOA or RCMOA previously in wells of cell culture plate and to allowing anti-native/denatured OAMAbs to these fixed antigens (NOA or RCMOA), was used. As liquid-phase method, a method comprising the steps of fixing rabbit anti-OA polyclonal antibodies in wells of cell culture plate, and allowing anti-native/denatured OAMAbs to these polyclonal antibodies while NOA or RCMOA are bound. Classes and subclasses of MAbs were determined according to Monoclonal mouse immunoglobulin isotyping kit (Pharmingen) as IgG1, IgG2a, IgG2b, IgG3, IgM, IgA, IgL ( $\kappa$ ) and IgL ( $\gamma$ ).

Please amend paragraph [0107] as follows:

[0107]

## 2) Combination conditions

Combinations of MAbs for detecting NOA or MAbs for detecting RCMOA were selected from the point of view of detection sensitivity by sandwich ELISA. As a result, 301B5 and 316G1, or 304E4(PNOA1; FERM ABP-10265) and 306B2 (PNOA2, FERM ABP-10266) were selected for NOA, and 117F9 and 119D11, or 948G11 (PDOA1; FERM ABP-10275) and 962B8 (PDOA2; FERM ABP-10276) were selected for RCMOA as combinations with high detection sensitivity.

~~Should the following examples be rewritten from 301B5 and 316G1/117F9 and 119D11 to 304E4 and 306B2/948G11 and 962B8.~~

Please amend paragraph [0109] as follows:

[0109]

[Table 10]

Test No.	Coating MAbs	Antigens	Secondary antibodies
Test 1	301B5	Serial dilution A (native)	Mixture of 316G1 and 117F9
	119D11		
	Mixture of 301B5 and 119D11		
Test 2	301B5	Serial dilution B (denatured)	
	119D11		
	Mixture of 301B5 and 119D11		
Test 3	301B5	Serial dilution C (native + denatured)	
	119D11		
	Mixture of 301B5 and 119D11		

Please amend paragraph [0131] as follows:

[0131]

### 5-2 Results

With the combination of PNOM1 and gold colloid labeled PNOM2, albumen solution which has been treated for 1 hour at room temperature and 50°C could be detected up to 10 ppb. Further, albumen which has been treated for 1 hour at 75°C and 100°C, could be detected up to 100ppb. From this result, for foods which have been subjected to heat-treatment corresponding to a treatment for 1 hour at 100°C, even by not using a denaturant such as urea, albumen could be detected up to 100ppb by a simple extraction by using an immunochromato strip of this anti-[[OMMab]] OMMAB. However, as detection was not possible with a heat treatment exceeding 100°C by an immunochromatography of OM, a solubilized treatment by urea, such as in the above, was necessary.